

CLAIMS

What is claimed is:

- 5
1. A method for obtaining a transplastomic plant comprising the steps of:
- 10 a) introducing into a plant cell plastid a first recombinant nucleic acid construct comprising a promoter functional in a plant cell plastid, a nucleic acid sequence encoding a protein providing for tolerance to a plastid lethal compound and a transcriptional termination region functional in a plant cell plastid and a second recombinant nucleic acid construct comprising a promoter functional in a plant cell plastid, a nucleic acid sequence encoding a protein providing for tolerance to a plastid non-lethal compound and a transcriptional termination region functional in a plant cell plastid,
- 15 b) placing said plant cell having said first and second nucleic acid constructs on a first culture medium comprising a plastid non-lethal compound for a period of time sufficient to permit said plant cell to replicate, and
- 20 c) placing said plant cell on a second culture medium comprising a plastid lethal compound for a period of time sufficient to select plant cells capable of growing in the presence of said plastid lethal compound.
2. The method according to Claim 1, further comprising:
- d) obtaining a transplastomic plant having only said first construct.
- 25 3. The method according to Claim 1, wherein said nucleic acid sequence encodes for proteins selected from the group consisting of genes providing tolerance to herbicides, inhibitors of plastid metabolic pathways, proteases, and nucleases.
- 30 4. The method according to Claim 3, wherein said nucleic acid sequence encodes for proteins selected from the group consisting of 5-enolpyruvylshikimate-3-phosphate synthase, nitrilases, phosphinothricin acyltransferase, and acetohydroxyacid synthase.

5. The method according to Claim 1, wherein said plastid non-lethal compound is selected from the group consisting of kanamycin, spectinomycin, streptomycin, paromomycin, and lincomycin.

5

6. The method according to Claim 1, wherein said plastid lethal compound is selected from the group consisting of herbicides, protease, nuclease, and a plastid metabolic pathway inhibitor.

10

7. The method according to Claim 1, wherein said plastid lethal compound is selected from the group consisting of glyphosate, phosphinothricin, norfluorazone, and atrazine.

15

8. The method according to Claim 1, wherein said first and said second constructs further comprise regions of plastid homology.

20

9. The method according to Claim 1, wherein said plant cell is selected on said first culture medium for less than about 12 weeks.

10. The method according to Claim 1, wherein said plant cell is selected on said first culture medium for less than about 8 weeks.

25

11. The method according to Claim 1, wherein said plant cell is selected on said first culture medium for less than about 4 weeks.

12. The method according to Claim 1, wherein said plant cell is selected on said first culture medium for less than about 3 weeks.

30

13. A method for obtaining a transplastomic plant comprising the steps of:

- a) introducing into a plant cell plastid a recombinant nucleic acid construct comprising a promoter functional in a plant cell plastid, a nucleic acid sequence encoding a protein providing for tolerance to a plastid lethal

compound and a transcriptional termination region functional in a plant cell plastid,

- 5 b) placing said plant cell having said nucleic acid construct on a first culture medium comprising a sublethal amount of a plastid lethal compound for a period of time sufficient to permit said plant cell to replicate, and
- c) placing said plant cell on a second culture medium comprising a plastid lethal compound for a period of time sufficient to select plant cells capable of growing in the presence of said plastid lethal compound.

10 14. The method according to Claim 13, wherein said nucleic acid sequence encodes for proteins selected from the group consisting of genes providing tolerance to herbicides, inhibitors of plastid metabolic pathways, proteases, and nucleases.

15 15. The method according to Claim 14, wherein said nucleic acid sequence encodes for proteins selected from the group consisting of 5-enolpyruvylshikimate-3-phosphate synthase, nitrilases, phosphinothricin acyltransferase, and acetohydroxyacid synthase.

20 16. The method according to Claim 13, wherein said plastid lethal compound is selected from the group consisting of herbicides, protease, nuclease, and a plastid metabolic pathway inhibitor.

25 17. The method according to Claim 13, wherein said plastid lethal compound is selected from the group consisting of glyphosate, phosphinothricin, norfluorazone, and atrazine.

 18. The method according to Claim 13, wherein said nucleic acid construct further comprise regions of plastid homology.

30 19. The method according to Claim 13, wherein said plant cell is selected on said first culture medium for less than about 12 weeks.

20. The method according to Claim 13, wherein said plant cell is selected on said first culture medium for less than about 8 weeks.

21. The method according to Claim 13, wherein said plant cell is selected on
5 said first culture medium for less than about 4 weeks.

22. The method according to Claim 13, wherein said plant cell is selected on said first culture medium for less than about 3 weeks.

10

15
FOIA b 7 - DCE 4060